Microbial Diagnostics Exercise: Background and Mission

Background

P. aeruginosa is an opportunistic pathogen that causes a range of acute diseases in both humans and animals due to its large number of virulence genes which directly interfere with different hosts and their defence systems. One of the best studied human infections is airway infections in patients suffering from the genetic disorder cystic fibrosis (CF). Due to a severely reduced mechanical clearing of the airways in these patients they acquire multiple infections of various bacteria and fungi, but the major cause of morbidity and mortality is chronic infection of *P. aeruginosa* in the lungs.

The infection process of *P. aeruginosa* in the CF airways is associated with extensive genetic adaptation and microevolution of the infecting bacteria. The accumulation of mutations results in genetic variants with phenotypes of which many are not usually observed among environmental isolates. These phenotypes include loss of motility (swim/twitch/swarm) and increased antibiotic resistance. In some cases, specific genes have been found to be hot spots for mutations during CF infections. These common targets include *lasR*, which encodes a quorum-sensing regulator, and *mucA*, which results in the overproduction of alginate and conversion to the frequently found mucoid phenotype.

Mission

You have been given 10 isolates of *P. aeruginosa* from a cystic fibrosis patient X. The isolates have been sampled at the same time-point, but from different body sites. It is now your task (1) to determine if the 10 isolates belong to the same clone type (AT chip genotyping), (2) and to determine the resistance profile for all the isolates (disk diffusion assay).

It will be important for the doctors to know whether it is a monoclonal infection (*e.i.* all the isolates belong to the same clone type) and whether isolates from the different body sites should be treated differently (*e.i.* does the different isolates have similar antibiotic resistance profiles).

Also, you are provided with additional information about the isolates. For example whether they are mucoid (overproduction of alginate). How does this information correlate with your observed clone types and resistance profiles?

Relevant references

Jelsbak L, Johansen HK, Frost A, Thøgersen R, Thomsen LE, Ciofu O, Yang L, Haagensen J, Høiby N, Molin S. 2007. Molecular epidemiology and dynamics of *Pseudomonas aeruginosa* populations in lungs of cystic fibrosis patients. Infect
Immun. 75(5):2214-24.

Yang L, Jelsbak, L, Marvig RL, Damkiær S, Workman CT, Rau MH, Hansen SK, Folkesson A, Johansen HK, Ciofu, O, Høiby N, Sommer MO, and Molin S. 2011. Evolutionary dynamics of bacteria in a human host environment. Proc Natl Acad Sci USA. doi: 10.1073/pnas.1018249108

Wiehlmann L, Wagner G, Cramer N, Siebert B, Gudowius P, Morales G, Köhler T, van Delden C, Weinel C, Slickers P, Tümmler B. 2007. Population structure of Pseudomonas aeruginosa. Proc Natl Acad Sci USA. 104(19):8101-6.